

181SecondRevisedSeqList
SEQUENCE LISTING

<110> UMBI
Bryan, Philip N.

<120> Engineered Proteases for Affinity Purification and Processing of
Fusion Proteins

<130> 4115-181

<140> 10/567,073
<141> 2006-02-03

<150> US 60/493,032
<151> 2003-08-06

<150> PCT/US04/021049
<151> 2004-06-29

<160> 14

<170> PatentIn version 3.5

<210> 1
<211> 30
<212> PRT
<213> Bacillus amyloliquefaciens

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Met Arg Gly Lys Lys Val Trp Ile Ser Leu Leu Phe Ala Leu Ala Leu
1 5 10 15

Ile Phe Thr Met Ala Phe Gly Ser Thr Ser Ser Ala Gln Ala
20 25 30

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<213> Bacillus amyloliquefaciens

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Ala Gly Lys Ser Asn Gly Glu Lys Lys Tyr Ile Val Gly Phe Lys Gln
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Thr Met Ser Thr Met Ser Ala Ala Lys Lys Lys Asp Val Ile Ser Glu
20 25 30

Lys Gly Gly Lys Val Gln Lys Gln Phe Lys Tyr Val Asp Ala Ala Ser
35 40 45

Ala Thr Leu Asn Glu Lys Ala Val Lys Glu Leu Lys Lys Asp Pro Ser
50 55 60

Val Ala Tyr Val Glu Glu Asp His Val Ala His Ala Tyr
65 70 75

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<210> 3
 <211> 275
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 <213> Bacillus amyloliquefaciens

<400> 3

Ala Gln Ser Val Pro Tyr Gly Val Ser Gln Ile Lys Ala Pro Ala Leu
 1 5 10 15

His Ser Gln Gly Tyr Thr Gly Ser Asn Val Lys Val Ala Val Ile Asp
 20 25 30

Ser Gly Ile Asp Ser Ser His Pro Asp Leu Lys Val Ala Gly Gly Ala
 35 40 45

Ser Met Val Pro Ser Glu Thr Asn Pro Phe Gln Asp Asn Asn Ser His
 50 55 60

Gly Thr His Val Ala Gly Thr Val Ala Ala Leu Asn Asn Ser Ile Gly
 65 70 75 80

Val Leu Gly Val Ala Pro Ser Ala Ser Leu Tyr Ala Val Lys Val Leu
 85 90 95

Gly Ala Asp Gly Ser Gly Gln Tyr Ser Trp Ile Ile Asn Gly Ile Glu
 100 105 110

Trp Ala Ile Ala Asn Asn Met Asp Val Ile Asn Met Ser Leu Gly Gly
 115 120 125

Pro Ser Gly Ser Ala Ala Leu Lys Ala Ala Val Asp Lys Ala Val Ala
 130 135 140

Ser Gly Val Val Val Val Ala Ala Ala Gly Asn Glu Gly Thr Ser Gly
 145 150 155 160

Ser Ser Ser Thr Val Gly Tyr Pro Gly Lys Tyr Pro Ser Val Ile Ala
 165 170 175

Val Gly Ala Val Asp Ser Ser Asn Gln Arg Ala Ser Phe Ser Ser Val
 180 185 190

Gly Pro Glu Leu Asp Val Met Ala Pro Gly Val Ser Ile Gln Ser Thr
 195 200 205

Leu Pro Gly Asn Lys Tyr Gly Ala Tyr Asn Gly Thr Ser Met Ala Ser
 210 215 220

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Pro His Val Ala Gly Ala Ala Ala Leu Ile Leu Ser Lys His Pro Asn
225 230 235 240

Trp Thr Asn Thr Gln Val Arg Ser Ser Leu Glu Asn Thr Thr Thr Lys
245 250 255

Leu Gly Asp Ser Phe Tyr Tyr Gly Lys Gly Leu Ile Asn Val Gln Ala
260 265 270

Ala Ala Gln
275

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Ala Gly Lys Ser Asn Gly Glu Lys Lys Tyr Ile Val Gly Phe Lys Ser
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Gly Ile Lys Ser Cys Ala Lys Lys Gln Asp Val Ile Ser Glu Lys Gly
20 25 30

Gly Lys Leu Gln Lys Cys Phe Lys Tyr Val Asp Ala Ala Ser Ala Thr
35 40 45

Leu Asn Glu Lys Ala Val Lys Glu Leu Lys Lys Asp Pro Ser Val Ala
50 55 60

Tyr Val Glu Glu Asp Lys Val Ala Lys Ala Tyr
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<212> PRT
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<223> pr8FKAM variant of SEQ ID NO: 2

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Ala Gly Lys Ser Asn Gly Glu Lys Lys Tyr Ile Val Gly Phe Lys Ser
1 5 10 15

Gly Ile Lys Ser Cys Ala Lys Lys Gln Asp Val Ile Ser Glu Lys Gly
Page 3

Gly Lys Leu Gln Lys Cys Phe Lys Tyr Val Asp Ala Ala Ser Ala Thr
35 40 45

Leu Asn Glu Lys Ala Val Lys Glu Leu Lys Lys Asp Pro Ser Val Ala
50 55 60

Tyr Val Glu Glu Asp Lys Val Phe Lys Ala Met
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Gly Ile Lys Ser Cys Ala Lys Lys Gln Asp Val Ile Ser Glu Lys Gly
20 25 30

Gly Lys Leu Gln Lys Cys Phe Lys Tyr Val Asp Ala Ala Ser Ala Thr
35 40 45

Leu Asn Glu Lys Ala Val Lys Glu Leu Lys Lys Asp Pro Ser Val Ala
50 55 60

Tyr Val Glu Glu Asp Lys Val Phe Arg Ala Met
65 70 75

<210> 7
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<212> PRT
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<223> C-terminal portion of prodomain

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<221> VARIANT
<222> (6)..(6)
<223> Xaa is Phe or Tyr

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<221> VARIANT
<222> (9)..(9)
<223> Xaa is Met, Lys or Tyr

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<400> 7

Glu Glu Asp Lys Leu Xaa Gln Ser Xaa
1 5

<210> 8

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<212> PRT

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<223> Residues 16-21 of SEQ ID NO: 2

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Gln Thr Met Ser Thr Met
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<211> 4

<212> PRT

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<223> Substitution of residues 16-21 of subtilisin prodomain

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Ser Gly Ile Lys
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<210> 10

<211> 4

<212> PRT

<213> Artificial Sequence

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<223> Substitution of P1-P4 of subtilisin prodomain

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Phe Lys Ala Met
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<223> Substitution of P1-P4 of subtilisin prodomain

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Phe Lys Ala Tyr
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<220>

<223> Substitution of P1-P4 of subtilisin prodomain

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Phe Lys Ala Phe

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<210> 13

<211> 4

<212> PRT

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<223> P1-P4 of wild-type subtilisin prodomain

<400> 13

Ala His Ala Tyr

1

<210> 14

<211> 4

<212> PRT

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<223> Substitution of P1-P4 of subtilisin prodomain

<400> 14

Phe Arg Ala Met

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